EFFECT OF FEEDING RATE AND FREQUENCYON GROWTH PERFORMANCE, SEX CONVERSION RATIO AND PROFITABILITY OF NILE TILAPIA (*OREOCHROMIS NILOTICUS*) FRY IN HAPA AT COMMERCIAL HATCHERIES

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SUMMARY

This trial was conducted for twenty one days to evaluate the efficacy of feeding rate and frequency on growth performance, sex conversion ratio and profitability of monosex tilapia (*Oreochromis niloticus*) fry followed by a 45 days feeding trial to determine sex conversion ratio. A total of 360000 fry of three days- old (weight 10.99±0.01mg in average) were stocked in twelve hapa (2×4×1 m) at the rate of 30000 fry/hapa. The hapas were divided into six treatments on the basis of feeding rates and frequencies viz. changeable rates 30>20>10 and 4 times/day (T1), rate 30>20>10 and 6 times/day (T2), constant rate 20% for 4 times/day (T3), rate 20% for 6 times/day (T4), rate 15% for 4 times/day (T5) and rate 15% for 6 times/day (T6) having two replications. Fry were fed mixed feed with 17α-methyltestosterone (MT) hormone at a rate of 100 mg/kg diet and supplemented with mixture of vitamin C and selenium. The water quality parameters were within suitable ranges for proper growth of *O. niloticus*. After ending the trial period, significant variations (P≤0.05) were observed in six treatments and T2 showed the best performance in case of all parameters studied such as final weight, weight gain, feed conversion ratio, specific growth rate, sex conversion ratio and profitability. The lowest values were obtained with T5 (feeding rate 15% and 4 times/day).

Keywords: Nile tilapia, feeding rate, feeding frequency, specific growth rate, profit index and sex reversal.

INTRODUCTION

Reduced production costs and maintaining the quality of sex conversion ratio considered to be the most important priorities of hatcheries

Sexual dimorphism is a significant factor in *Oreochromis niloticus* where males grow significantly faster, larger and more uniform in size than females (Manosroi *et al.*, 2004; Bwanika *et al.* 2007 and Mensah *et al.* 2013). Although tilapias in general are known for their aquaculture potential but their growth and other production traits are largely influenced by genetics (Ajiboye and Yakubu 2010). Early sexual maturity of this species is a well-recognized problem. Use of monosex (all-male) has been widely, if inconsistently, promoted and adopted (Green *et al.*, 1997).

Synthetic androgens are used in fish culture as sex controlling agents and as growth promoters if energy is shut away from developing ovaries towards growth of somatic tissues (Rizkalla *et al.*, 2004). The more common method of generating mostly male populations is through the use of steroids fed to sexually undifferentiated fry, when the newly hatched tilapias are still developing their gonads. Even though they are determined genotypically, their phenotype or morphological characteristics can still be altered. By exposing the fish to forms of testosterone or estrogen, the gonad can switch (Al-Hakim *et al.*, 2012).

Although monosex male population can be obtained by direct or indirect methods, oral administration of *Oreochromis niloticus* has been reported to be the most preferred method in commercial uses (Green and Teichert-Coddington, 2000; Wahby and Shalaby, 2010; Celik *et al.*, 2011). 17 α -methyltestosterone (17 α -MT) is a synthetic male hormone which closely mimics the naturally-produced hormone testosterone. The most common sex-reversal treatment involves giving a powdered fish feed to the first-feeding (and still sexually undifferentiated) tilapia fry and diet contains 60-100 mg 17a-MT/kg of feed until the 28th days post hatching (Al-Hakim *et al.*, 2012).

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The relationship between rate of feeding and rate of conversion is very important in fish culture when fish are fed insufficiently or excessively, their growth or feed efficiency may decrease, resulting in increasing production costs and water quality deterioration (Winnie 2012). If fish are fed at too high rate, much of the food is wasted, and, even though growth may be excellent, the cost of production is excessive. Further, in some cases uneaten food decaying in the water may be detrimental to fish growth. If the feeding rate is too low, approaching the rate required for maintenance, growth is very slow, and the rate of conversion is very high (Shell 1996). Increased feeding frequencies decrease aggressive behavior in some fish species, these results in faster growth and less size variation. However, there is a limit to the frequency that will result in benefits. Because of their rapid growth, high energy requirements, and small stomachs, fry require frequent feeding. Fry should be fed as many as 8-10 times a day (Riche and Girling 2003). Thus the diet amount fed each time, or feeding frequency, may influence diet utilization. This is due to the fact that diet is directly applied to water and the non-untaken portion will be dissolved and lixiviated. Feed conversion ratio increase and environmental pollution are the results.

Since that the main aim is that fries uptake a high daily diet ratio to meet their nutrition requirements and thus ingest adequate hormonal amounts, and since that the high feeding frequency results in high daily diet intake ratio and small amounts of diet per feeding (Meer *et al.*, 1997), a higher frequency may be the most adequate, particularly in the sex reversal period of tilapia larvae culture. There are a wide variation in opinions about feeding frequency diverge from 2 times a day, as used by Guerrero (1975) pioneer experiments, 2 to 4 times a day as recommended by Popma and Green (1990), three times a day as used by Alcazar (1988), four times a day as used by Phelps *et al.* (1995) and by Vera Cruz and Mair (1994), 6 to 8 times a day as reported by Carberry and Hanley (1997), up to the minimum of 8 times a day as recommended by Lim (1997). With regard to the economic aspect, a higher feeding frequency would only be justified when absolutely necessary for a better performance. Needless to say, a higher feeding frequency would require higher costs with salary or with automatic feeders. Taking into account the above biological and economic aspects of sex reverted tilapia fries production, the aim of this experiment was to verify the minimal feeding frequency required for an optimal performance in that period.

The aim of this work was to investigate the effect of fry feeding rate and frequency on growth performance, sex reversaland profitability of *O. niloticus* fry in hapa pond system at commercial hatcheries.

MATERIALS AND METHODS

This study was conducted in a greenhouses commercial hatchery at Edko province, Beheira Governorate, Egypt to evaluate the effect of feeding rate and frequency on growth performance, sex conversion ratio and profitability of Nile tilapia fry. The experiment began in 15/03/2015 and lasted for 21 days (basic trials) followed by a 45 days feeding experiment to determine sex conversion ratio.

Three hundred and sixty thousand fry (yolk sac) were equally divided on twelve hapa located in fry earthen pond. Each hapa has an area of 2×4 m with a water depth of 80-85 cm. fry stocked at a rate of 5000/m³ with an average weight of 10.99 ± 0.01 mg/fry.

The first replicate of all treatments started in a day and the second replicate was started in the second day because the hatchery production of fry was not enough to supply enough fries to start all treatments in the same day

Diets: fries in Hapas were fed on a diet that contained 53% protein (80% fish meal herring mixed with 20% flour) with 17α methyl testosterone at a rate of 100 mg/kg diet(Al-Hakim *et al.*, 2012) and supplemented with mixture of vitamin C and selenium. Feed was offered at hours 8.0, 11.0, 14.0, and 17.0 for treatment 4times/day and at hour 8.0, 10.0, 12.0, 14.0, 16.0 and 18.0 for treatment 6times/day as follows:

 $-T_1$ feed fries at changeable feeding rate 30% of body weight in first week then decreased to 20% in the second week and 10% in the third week 4 times/day.

 $-T_2$ feed fries at changeable feeding rate 30% of body weight in first week then decreased to 20% in the second week and 10% in the third week 6 times/day.

-T₃ feed fries at constant feeding rate 20% of body weight for 21 days 4 times/day.

-T₄ feed fries at constant feeding rate 20% of body weight for 21 days 6 times/day.

-T₅ feed fries at constant feeding rate 15% of body weight for 21 days 4 times/day.

-T₆ feed fries at constant feeding rate 15% of body weight for 21 days 6 times/day.

Determination of sex conversion:

At the end of the experiment, five hundred fries from each treatment were confined in small hapa $(1 \times 2 \text{ m})$ and fed for 45 days until fry weight reached 4-5 g on artificial diet containing 30% CP to determine sex conversion ratio. The identification of the phenotypic sex for 200 juvenile fry from each treatment was determined by microscopic examination of the gonads. The thin gonad (thread-like structure lies along the dorsal side of the abdominal cavity) was extracted very carefully, placed on a glass slide and stained with a drop of aceto-carmine stain then it was lightly squashed with a glass cover slip and examined at 10X magnification for the identification of the juvenile gonads. The fish was a presumptive male and female if densely packed oocytes were found as reported by Guerrero and Shelton (1974).

Water temperature, pH, dissolved oxygen (DO₂), ammonia (NH₃-N), nitrate and salinity throughout the experimental period were measured periodically in the morning and at noon by centigrade thermometer, Orion digital pH meter model 201, oxygen meter, Cole Parmer model 5946, HACH test kit ammonia mid-range 0-3 mg/L model NI-8, HACH test kit Nitrate/Nitrite model NI-12 and TDS apparatus, respectively.

Parameters:

At the end of the experiment, growth parameters and survival rate were measured as follows:

- Weight gain = Final weight Initial weight (Effiong et al., 2009).
- Daily gain = Weight gain, g /period in days. (Effiong et al., 2009).
- *Specific growth rate (SGR, %)* = 100 (ln Final weight-ln Initial weight)/period in days, where ln is the natural log. (Effiong *et al.*, 2009).
- *Feed conversion ratio (FCR)* = feed offered / weight gain (Effiong *et al.*, 2009).
- Survival rate (SR) %= Final number of fish /Initial number of fish x 100.(Charo-Karisa et al., 2006)

Economic analysis

A simple economic analysis was used to assess the cost effectiveness of diets used in the feed trial. The cost of feed was calculated using market prices, taking into consideration the cost of feed and the transport fare with the assumption that all other operating costs remained constant (e.g. cost of constructing hapa, cost of fingerlings and labor). Indices for economic evaluation included:

(i) Incidence cost (IC), which was calculated as:

IC= Cost of feed/ No. of fry produced

IC is actually the cost of feed to produce 1000 fry (relative cost per unit), and the lower the value, the more profitable using that particular feed (Nwanna, 2003; Abu *et al.*, 2010)

- (ii) Profit index (PI), which was calculated as:
- PI = value of fish produced/ Cost of feed

Statistical Analysis:

Data were statistically analyzed using a one-way analysis of variance using SPSS version 16 (2007). Mean of treatments were compared by Duncan Multiple Range Test (1955) when the differences were significant.

RESULTS AND DISCUSSION

Water quality parameters:

Water quality parameters were measured during the study in fish hapas. Mean values of water quality parameters were recorded as follows: dissolved oxygen (DO) 5.8 ± 0.13 mg/L, pH 8.12 ± 0.14 , temperature 29 ± 1.0 °C, NH₃-N 0.4 ± 0.15 mg/L and salinity 2.6 ± 0.18 %o respectively. These values are within the safe ranges and acceptable for the spawning and growth of tilapia as reported by El-Sayed (2006), Magid and Babiker (1975), Ross (2000) and El-Sherif and El-Feky (2008).

Effect of feeding rate on growth performance of Nile tilapia fry:

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Table (1) illustrated that the feeding rate significantly ($P \le 0.05$) affected the growth performance parameters, final weight (FW), weight gain (WG), daily gain (DG), specific growth rate (SGR) and feed conversion ratio (FCR). The treatment group which received 20% feeding rate showed the best final weight (162.93 mg), weight gain (151.95 mg), daily gain (7.23 mg) and SGR (12.84) followed by the treatment group received changeable feeding rate 30>20>10 which were 145.39, 134.41, 6.04 and 12.29 for FW, WG, DG and SGR respectively and these parameters are insignificant ($P \ge 0.05$) between changeable feeding rate 15%.

Feeding	Initial	Final	Weight	Daily	SGR, %	Survival	FCR
rate	weight, mg	weight, mg	gain, mg	gain, mg	50K, %	rate, %	
30>20>10	10.98	145.39 ^b	134.41 ^b	6.40 ^b	12.29 ^b	90.87ª	1.18 ^b
20	10.98	162.93ª	151.95ª	7.23 ^a	12.84 ^a	90.28 ^a	1.46 ^a
15	10.99	139.49 ^b	128.50 ^b	6.12 ^b	12.10 ^b	80.95 ^b	1.45 ^a
SED	0.01	5.39	5.39	0.26	0.17	2.51	0.08

Table (1). Effect of feeding rate on growth performance of Nile tilapia fry during sex reversal

* Average in the same column having different superscripts significantly different at ($P \le 0.05$).

** SED, standard error of a difference between 2 means = $\sqrt{(2 \times Error MS/r)}$

Survival rate and FCR values showed a significant differences ($P \le 0.05$) between feeding rate treatment group. The results may be due to the large quantity of feed offered to fry in the first week of feeding with changeable feeding 30>20>10 and 20% feeding rate these lead to fry consumed more feed than that of feeding rate 15% which were low quantity of feed and large, strong fry consumed feed only, but the weak fry don't crowded resulting increased mortality and lowest growth of fries feeding 15%.

The poor FCR noticed with feeding rate 20% may be attributed to the presence of more feed than fry requirements specially in the third week but changeable feeding 30>20>10 during sex reversal period at 30% in the first week then decreased to 10% in the third week improved FCR due to the low quantity of feed. These results are in agreement with the results obtained by Santiago *et al.* (1987) and El-Sayed (2002) who reported that fish growth rates and survival were extremely poor at 10% feeding level, and improved significantly with increasing feeding levels up to 30%, and levelled off with further increase in feeding levels.

Effect of feeding rate on sex conversion ratio and profitability of Nile tilapia fry

The effect of feeding rate on sex conversion ratio showed a significant ($P \le 0.05$) differences between feeding rates and the best sex conversion ratio obtained with changeable feeding rates 30>20>10 (96.75% male) and feeding 20% (95.75% male) but the feeding rate 15% resulted the lowest male % (93%) (Table 2). The results may be explained on the basis that changeable feeding rate and 20% feeding rate provides a large quantity of feed offered to fry specially in the first week which sex conversion happed compared to the small quantity offered by the 15% feeding rate, the larger tilapia dominate the area around the feeder and consume most of the feed, resulting in considerable size variation and often poor sex reversal. Pandian and Varadaraj (1987), Phelps and Popma (2000) distained similar results on poor sex reversal suing similar feeding rate.

The calculation of profitability showed that the treatment group that received changeable feeding rate 30>20>10 was more profitability than the other two treatment groups. The results of this particular work recommended the use of changeable feeding rate to get the highest profit index (9.92) and highest male percent (96.75).

Table (2).	Effect of feeding rate	on sex reversal and	d profitability	of Nile tilapia fry

Feeding rate, %	Male, %	Feed offered	Feed cost	Fry No sales (V)	Values of fry sales	Profit index
30>20>10	96.75 ^a	4478.75°	72.11 ^c	28625ª	716 ^a	9.92ª
20	95.75ª	6227.50 ^a	100.26 ^a	28438ª	711 ^a	7.09 ^c
15	93.00 ^b	4645.00 ^b	74.78^{b}	25500 ^b	638 ^b	8.53 ^b
SED	0.62	29.80	0.48	790	19.74	0.27

* Average in the same column having different superscripts significantly different at ($P \le 0.05$).

** SED, standard error of a difference between 2 means = $\sqrt{(2 \times Error MS/r)}$

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Effect of feeding frequency on growth performance of Nile tilapia fry:

Results in Table (3) revealed that the growth performance was insignificantly (P \geq 0.05) affected in all parameters such as final weight, weight gain, daily gain, specific growth rate and feed conversion ratio. The results take a constant trend with feeding frequency 4 or 6 times/day and the best with feeding frequency 6 times/day. These results are in agreement with that obtained by Ferdous *et al.* (2014), Luthada and Jerling (2013), Pouomogne and Ombredane (2001) and Sanches and Hayashi (2001).

Feed frequency	Initial weight	Final weight	Weight gain	SGR	Survival rate	FCR	Profit index
4	10.99	146.14	135.15	12.31	85.85	1.40	8.35
6	10.99	152.39	141.40	12.51	88.89	1.29	8.68
SED	0.011	11.55	11.56	0.36	5.26	0.14	1.28

Table (3). Effect of feed frequency on growth performance and profit index of Nile tilapia fry

** SED, standard error of a difference between 2 means = $\sqrt{(2 \times Error MS/r)}$

Effect of feeding rate and frequency on growth performance of Nile tilapia fry:

Table (4) showed that the growth performance was significantly (P ≤ 0.05) affected in all parameters such as final weight, weight gain, daily gain, specific growth rate and feed conversion ratio. The results obtained with feeding rate 20% and feeding 6 times/day showed the best final weight (166.73 mg), weight gain (155.74 mg), daily gain (7.42 mg) and SGR (12.95) and the lowest values were recorded with 15% feeding rate and 4 times/day, but survival rate and FCR were the best with changeable feeding rate at 6 times/day (92.06 and 1.13) than the other treatments. The lowest values were obtained with 15% feeding rate and 4 times/day. The results of this particular work are clearly supported by the findings of Ferdous *et al.* 2014 and Pouomogne and Ombredane. 2001 who stated that, increasing the frequency of feeding in tilapia fry positively correlated with better fishgrowth performance. Also, Luthada and Jerling (2013) showed that weight gain and specific growth rate at higher feeding frequencies have also been reported for red tilapia hybrid fry by Siraj *et al.*, (1988) and juvenile *O. niloticus* Riche *et al.* (2004).

Sena and Trevor (1995) suggested that the manual feeding frequency severaltimes per day is the most appropriate for intensive grown tilapia. Tung and Shiau (1991) also confirmed that weight gain of *O*. *niloticus* increased with increasing feeding frequency.

Treatment	Initial	Final	Weight	Daily	SGR	Survival	FCR
	weight	weight	gain	gain	SOK	rate	
1	10.99	143.17 ^c	132.18 ^c	6.29 ^c	12.22 ^c	89.68 ^a	1.20 ^c
2	10.99	147.62 ^{bc}	136.63 ^{bc}	6.51 ^{bc}	12.37 ^{bc}	92.06 ^a	1.13°
3	10.99	159.13 ^{ab}	148.14^{ab}	7.05 ^{ab}	12.73 ^{bc}	89.29ª	1.49 ^a
4	10.99	166.73 ^a	155.74 ^a	7.42^{a}	12.95 ^a	91.27 ^a	1.40 ^b
5	10.99	136.14 ^c	125.15°	5.96°	11.98°	78.57 ^b	1.51 ^a
6	10.99	142.83°	131.84 ^c	6.28 ^c	12.21°	83.33 ^b	1.34 ^b
SED	0.01	4.81	4.81	0.23	0.15	2.00	0.03

Table (4). Effect of feeding rateand frequency on growth performance of Nile tilapia fry

* Average in the same column having different superscripts significantly different at (P ≤ 0.05). ** SED, standard error of a difference between 2 means= $\sqrt{(2 \times \text{Error MS/r})}$

Effect of feeding rate and frequency on sex conversion ratio and profitability of Nile tilapia fry

The results of feeding rate and frequency on sex conversion ratio (Table 5) showed a significant ($P \le 0.05$) differences between treatments and the best sex conversion ratio obtained with changeable feeding 30 > 20 > 10 and feeding 6 times/day while the feeding rate 15% and feeding 4 times /day recorded the lowest male %. The results may be due to a large quantity of feed offered to fry specially in the first week which sex conversion, take place, compared to rate 15% and 4 times/day which weak fry don't consume enough feed resulting poor sex reversal (92%). When small quantities of feed are released uniformly throughout the day, the larger tilapia dominate the area around the feeder and consume most of the feed, resulting in considerable size variation and often poor sex reversal.

Treatment	Male, %	Feed offered, g	Feed cost	Fry No sales (V)	Values of fry sales	Profit index
1	96.50 ^{ab}	4487.50 ^c	72.25°	28250 ^a	706.25 ^a	9.77ª
2	97.00^{a}	4470.00 ^c	71.97°	29000 ^a	725.00 ^a	10.07^{a}
3	95.00 ^b	6210.00 ^a	99.98ª	28125 ^a	703.13 ^a	7.03 ^d
4	96.00 ^{ab}	6245.00 ^a	100.54 ^a	28750 ^a	718.75 ^a	7.15 ^d
5	92.50°	4665.00 ^b	75.11 ^b	24750 ^b	618.75 ^b	8.24 ^c
6	93.50°	4625.00 ^b	74.46 ^b	26250 ^b	656.25 ^b	8.81 ^b
SED	0.58	28.47	0.46	633.28	15.83	0.20

Table (5). Effect of feeding rate and frequency onsex reversal andprofitability of Nile tilapia fry

* Average in the same column having different superscripts significantly different at (P \leq 0.05). ** SED, standard error of a difference between 2 means= $\sqrt{(2 \times Error MS/r)}$

Regarding the profitability of hatchery changeable feeding 30>20>10 with feeding 6 times/day was the best than other treatment. The results of this research work revealed that changeable feeding 30>20>10 with feeding 6 times/day produced best male % (97) and profit index (10.07) followed by changeable feeding 30>20>10 with feeding 4 times/day produced male % (96.50) and profit index was (9.77) but the lowest male % with feeding rate 15% with feeding 4 times/day.

CONCLUSION

The first days after hatching are crucial in the process of sex reversal. Diets containing 17α methyltestosterone hormone should be given in sufficient quantity. The optimal feeding rate and frequency can lead to maximization of profit. In this particular study it was found that changeable feeding rate 30>20>10 with a frequency of 6 times / day was the proper.

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تأثير معدل التغذية وعدد مراتها على مظاهر النمو ونسبة التحول الجنسى والربحية لزريعة البلطى النيلى في الهابات بالمفرخات التجارية

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أجريت هذه التجربة لمدة واحد وعشرين يوما لتقييم تأثير معدل التغذية وعدد مراتها على أداء النمو، ونسبة تحويل الجنس وربحية زريعة البلطى النيلى وحيد الجنس واتبعت ب 45 يوما تجربة تغذية لتقدير نسبة التحول الجنسى. وزع 30000 زريعة عمر ثلاثة ايام مقوسط وزنها (10.9 ± 10.00 في اثني عشر هابا (2 × 4 × 1 م) بمعدل 30000 زريعة / هابا. تم تقسيم الهابات إلى ستة معاملات على أساس معدل التغذية وعدد مراتها. معدل تغذية متغير 2000 في 10.00 في اثني عشر هابا (2 × 4 × 1 م) بمعدل 30000 زريعة / هابا. تم تقسيم الهابات إلى ستة معاملات على أساس معدل التغذية وعدد مراتها. معدل تغذية متغير 2000 او 4 مرات / يوم(11) ، ومعدل 2000 او 6 مرات / يوم(12) ، ومعدل 2000 في الثني عشر هابار 2000 في الذي معدل تغذية متغير 2000 معاملات على أساس معدل التغذية وعدد مراتها. معدل تغذية متغير 2000 معدل 2000 أو 6 مرات / يوم(12) ، ومعدل 2000 في مرات / يوم(12) ، ومعدل 2000 في مرات / يوم(12) ، ومعدل 10.00 في مرات / يوم(12) ، ومعدل 2000 في مرات / يوم(12) ، معدل 10.00 في معاملة في هابتين. تم تغذية اليرقات بالعليقة مع 17 الفا ميثيل تستوستيرون كما مرات / يوم(12) والسيلينيوم. تم تقدير نوعية المياه وكانت مناسبة النمو السليم لزريعة البلطي النيلي. بعد انتهاء الفترة التجريبية، تم تدعيم العليقة بفيتامين 2000 والسليم لزريعة البلطي النيلي. بعد انتهاء الفترة التجريبية، تم تدعيم العليقة بفيتامين 2 والسيلينيوم. تم تقدير نوعية المياه وكانت مناسبة للنمو السليم لزريعة البلطي النيلي. بعد انتهاء الفترة التجريبية، تم تدعيم العليقة بفيتامين 2 والسيلينيوم. تم تقدير نوعية المياه وكانت مناسبة النمو السليم لزريعة البلطي النيلي. بعد انتهاء الفترة التجريبية، تم تدعيم العليقة بفيتامين 2 والليه الفوري في والته والسليم لزريعة المائم الزريعة البليني الوزن، النهائي، زيادة الوزن، ونمسبة التحويل الخاني، معدل النمو الوزن، ونمائلة والفيري والنه، ويا المائلة، والونه والونه، والعن والول النهائي، معدل النهائي، زيادة والوزن، والمول على أداء في مع عرم العاني، معدل النهائي، زيادة 2000 والوريم. والمول على أداء في معيع المعاملات وأطهرت 12 في مرات / يوم).